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ANTHOCHLOR PIGMENTS AND POLLINATION BIOLOGY. I. THE UV ABSORPTION OF *ANTIRRHINUM MAJUS* FLOWERS

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INTRODUCTION

The existence of UV-absorbing, floral pigmentation patterns invisible to the human eye, but visible to insect pollinators, was photographically documented early in this century (Richtmyer, 1923; Lutz, 1924). Identification of the chemical compounds responsible for producing these patterns was accomplished only recently when Thompson et al. (1972) demonstrated that flavonol glycosides produced the floral UV-absorption pattern present in *Rudbeckia hirta* L. Those workers noted (their footnote 16) that the anthochlor pigments (aurones and chalcones) also possess absorption maxima appropriate for participation in floral UV-absorption phenomena. This paper presents the first experimental demonstration of the involvement of aurones in imparting UV absorption to a flower.

MATERIALS AND METHODS

Bedding plants of a yellow, garden variety (unknown genetic stock) of *Antirrhinum majus* L. (Scrophulariaceae) were purchased from a local nursery supplier. Flowers were extracted with acidified MeOH. The yellow extract was concentrated under reduced pressure, streaked on Whatman 3MM paper, and chromatographed in one dimension using *n*-butanol, acetic acid (HOAc), water: 4, 1, 5 (upper phase, BAW). Compounds from the four bands detectable under UV illumination were eluted in MeOH, concentrated, and their spectral and chromatographic properties determined by the methods of Mabry, Markham, and Thomas (1970).

The following chromatographic solvents were used: various percentages of aqueous HOAc, BEW (*n*-butanol, ethanol, water: 4, 1, 2.2), TBA (tertiary-butyl alcohol, HOAc, water: 3, 1, 1) and Forestal's (HOAc, conc. HCl, water: 30, 3, 10).

Flowers and chromatograms were photographed in sunlight through a Kodak-Wratten 18A filter (Tiffen Optical Co., New York) to detect UV absorption. To detect the presence of anthochlor pigments, the flowers were exposed for ca. 10 min to fumes of conc. NH_4OH in a closed container.

TABLE 1. *Chromatographic and spectral properties of flavonoid pigments from* Antirrhinum majus *petals.*

Spot number	Identity	Color in UV light ($+NH_3$)[a]	R_f values (×100) in: BAW	BEW	TBA	H_2O	15% HOAc	30% HOAc	Spectral maxima in MeOH (nm)
1	Bracteatin-6-0-glucoside	Y(R-O)	10	23	05	00	03	05	264,323,409
2	Auresidin-6-0-glucoside	Y(R-O)	28	55	16	00	08	16	272,323,404
3	Luteolin-7-0-glucuronide, Chrysoeriol-7-0-glucuronide	Dk(Y)	39	46	31	08	25	37	256,268,349
4	Apigenin-7-0-glucuronide	Dk(Y)	57	57	43	11	30	53	271,332

[a] Color code: Dk = dark, R = red, O = orange, R = red, Y = yellow.

RESULTS

Flowers of the yellow-blossomed variety of the garden snapdragon, *Antirrhinum majus*, appear uniformly bright yellow (Sulfur Yellow, Horticultural Color Chart #1). Upon exposure to ammonia vapor a striking color change occurs which is characteristic of anthochlor pigments; the entire petal surface becomes uniformly bright orange (Burnt Orange, H.C.C. #o14). When photographed through appropriate UV-transmitting filters the entire petal surface appears as completely absorptive of UV-wavelength light. The pigments responsible for this absorption can be separated by paper chromatography and detected on paper by their in vitro UV absorption when chromatograms are photographed through appropriate filters. Four dark spots, consisting of five compounds, were noted on such photographs and the compounds were identified to be: 1) bracteatin-6-0-glucoside; 2) auresidin-6-0-glucoside; 3) a mixture of luteolin-7-0-glucuronide and chrysoeriol-7-0-glucuronide; and 4) apigenin-7-0-glucuronide. Chromatographic and spectral properties on which these identifications are based are given in Table 1.

DISCUSSION

The flavonoid constituents of petals of various varieties of *Antirrhinum majus* have been extensively studied by Jorgensen and Geissman (1955), Harborne (1963) and Gilbert (1973). My chemical analyses agree with the results of these earlier workers and suggest that one of the biological functions of the flavonoid compounds in the petals of the snapdragon is the attraction of pollinating insects. This is the first report of the participation by aurone glucosides and flavone glucuronides in the production of floral UV absorption. The existence of a UV absorption-reflection pattern due to the spatial segregation of anthochlor pigments has been noted by the author in flowers of other taxa (*Coreopsis* spp., *Bidens* spp.) and will be reported elsewhere. In contrast to these taxa, observations on cultivated *A. majus* show no UV patterning in that the entire petal appears UV absorptive. This situation may not obtain in the natural *A. majus* native to Mediterranean Europe. In the native plant the yellow pigmentation is restricted to the lip and only in cultivated forms has the pigmentation been spread by selective breeding from the lip to throughout the corolla. In the natural state the yellow, UV-absorbing lip may serve as an orientation cue to incoming pollinators. Samples of the native plant were unavailable to the author for investigation.

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